Dorsal root ganglion neurons maintained in a 3D culture model exhibit similar electrophysiological properties to fresh explants

Conference or Workshop Item

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INTRODUCTION: Tissue engineered culture models provide a powerful tool for neuroscience research[1]. They overcome limitations associated with monolayer cultures of neurons and glia by maintaining cells in a more realistic 3D spatial arrangement, and permit continuous monitoring and control of variables that cannot be achieved in animal models. Here we report the development of a system for recording electrophysiological behaviour in neurons in 3D culture.

METHODS: Dorsal root ganglia (DRGs) were harvested from adult rats, dissociated using collagenase, then seeded within 2 mg/ml Type I collagen gels and maintained in culture for 20 hours. Preparations were transferred to an interface recording chamber at 29 °C, perfused with culture medium at 150 µl/min and exposed to warmed and humidified oxygen (95 %) and carbon dioxide (5 %). Recordings were made using glass micropipettes filled with 3M KCl (electrode series resistance 60-80 MOhms) attached to an Axoclamp 2B amplifier and stored on a Macintosh computer. Neurons in 3D culture were compared to those in acutely hemi-sectioned DRG explants.

RESULTS: Resting membrane potential and input resistance were recorded from neurons in both 3D cultures and acutely hemi-sectioned control tissue. Characteristic membrane voltage responses to hyperpolarising current were obtained and injection of depolarising current elicited action potentials (Fig 1).

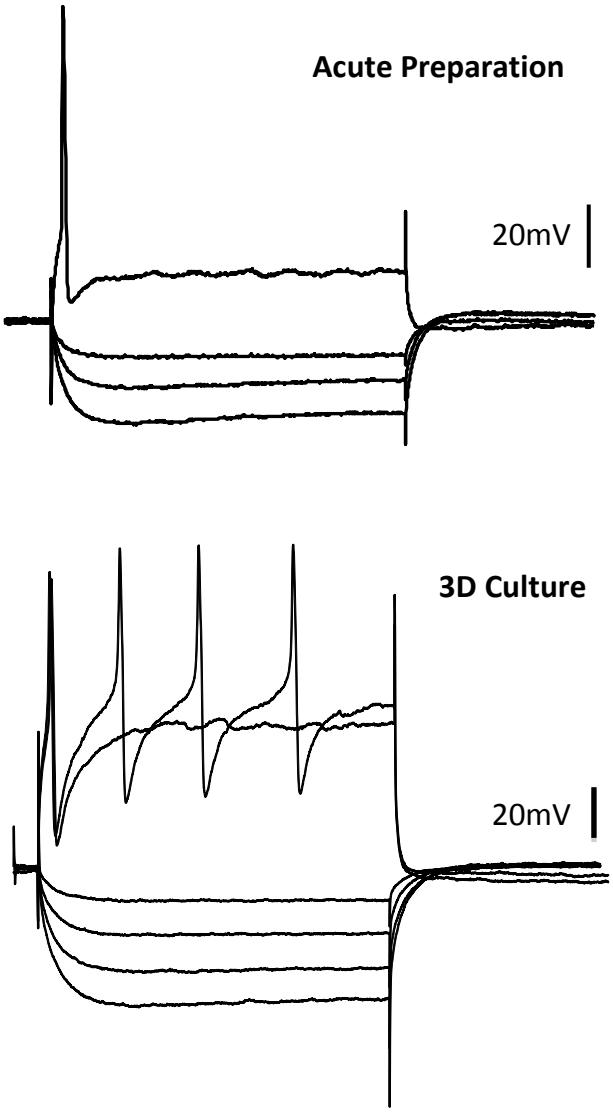

Fig 1:Upper Panel: traces recorded from a cell in an acutely hemi-sectioned DRG. The neuron had a membrane potential of -81 mV and input resistance of 110 MOhms. Traces show the membrane voltage responses to injections of hyperpolarising current (-0.1, -0.2 and -0.3 nA; 150 ms duration) and a depolarising current sufficient to elicit an action potential.

Lower panel: traces recorded from a DRG neuron in 3D culture. The neuron had a membrane potential of -87 mV and input resistance of 68 MOhms. Traces show the membrane voltage responses to injections of hyperpolarising current (-0.2, -0.4, -0.6 and -0.8 nA; 150 ms duration) and injections of depolarising current that elicited either a single action potential or a train of action potentials.

DISCUSSION & CONCLUSIONS: Adult rat DRG neurons maintained in 3D culture exhibit electrophysiological responses comparable to their counterparts in fresh tissue explants. This system provides a functional model in which neuronal responses can monitored. The reproducibility and control make this approach suitable for further development as a model for toxicity testing.

REFERENCES: [1] E. East & J.B.Phillips (2008) Tissue engineered cell culture models for nervous system research in *Tissue Engineering Research Trends* (ed G. N. Greco) Nova Science Publishers.